Recent reports described the differences in steam-volatile acidic and neutral substances in the leaves and smoke condensates of the four major cigarette types (Burdick *et al*, 1963; Burdick and Stedman, 1963; Schmeltz *et al*, 1963*a*, 1963*b*; Stedman *et al*, 1963). Also, the levels of such substances were determined in two series of flue-cured leaf samples designated "Aromatic" or "Aroma-deficient." It was concluded that no correlation was observed between leaf aromaticity and the level of total volatile neutrals (Burdick *et al*, 1963) but that aromatic samples showed higher levels of total, free volatile acids[2] than aroma-deficient samples (Schmeltz *et al*, 1963*a*, 1963*b*).

These conclusions were based on the assumption that all aromatic (or aroma-deficient) samples were equally aromatic (or aroma-deficient). Since publication of these data, the results of panel tests to determine the relative degree of aromaticity in these samples have been made available to us. Comparison of these panel test findings and the published chemical data has shown a tendency toward a relationship between the degree of aromaticity and the levels of total volatile acids and neutrals and has permitted some revision of our original conclusion. Details on the acids will be presented below and the findings on the neutrals will be described in a subsequent publication.

In an extension of this work, the total volatile acids have now been determined in smoke condensates of cigarettes made from the above two series of aromatic grades. The results of these determinations and their relationship with panel tests of the relative flavor of such cigarettes are described herein.

Experimental

The flue-cured tobaccos used in making the test cigarettes were those previously designated as Aromatic A-C and Aroma-deficient A-B (Burdick *et al*, 1963). The cigarettes were 70 mm in length and contained no filters or additives. The smoking machine and smoking conditions were identical with those previously described with one exception: the smoke collection system was modified by replacing the first in the series of spiral glass traps by a large cold finger containing a small amount of glass wool. The moisture content of the cigarettes was 12.2-12.9%. Forty-five cigarettes of each set were smoked for the analysis; approximately 55% of each cigarette was smoked and the actual % length smoked was determined by weight as previously discussed.

Certain changes were made in the previously used method of isolating the acids in order to improve the recovery of acetic acid. The acidic substances from the condensate of 45 cigarettes were obtained in an alkaline solution (0.5% aqueous NaOH) from which neutrals and bases were removed as previously described (Stedman *et al*, 1963). The alkaline solution (125 ml) was adjusted to pH 7.8 with H_2SO_4 and extracted with ether (3 times, 150 ml total) giving an ether solution of weak acids and an alkaline solution of strong acids. The pH of the latter was adjusted to 1.0 with H_2SO_4 and, after saturation with NaCl, was steam distilled. One liter of distillate was collected and, after saturation with NaCl, the distillate was extracted with ether (5 times, 750 ml total). The ether extract was dried over $MgSO_4$ and reduced in volume to 10.0 ml using the solvent removal procedure previously outlined; this concentrate was analyzed for volatile acids employing the diethylene glycol adipate polyester-phosphoric acid column, flame ionization detection and other conditions described by Schmeltz *et al* (1963*a*, 1963*b*). The reported levels are representative values obtained from several analyses of the samples.

Results and Discussion

The chromatograms of the volatile strong acidic fraction of all smoke condensates were qualitatively similar to that previously obtained for bright cigarettes (Schmeltz *et al*, 1963*b*). The above change in isolation procedure resulted in an increase in recovery of known acetic acid from 10-30% (Stedman *et al*, 1963) to about 60%. The latter is considered a satisfactory yield for acetic acid in standard methods for determining volatile acids which employ steam distillation (Association of Official Agricultural Chemists, Official Methods of Analysis, 1960). Formic acid is relatively undetectable by flame ionization and was not determined.

Table 1 shows the relative amounts of the acids in the various smoke condensates using Aromatic A as the reference. The three aromatic samples contained more isovaleric acid and components in uni

[1] *Eastern Utilization Research and Development Division, Agricultural Research Service, United States Department of Agriculture.*

[2] *The terms "total volatile neutrals" and "total volatile acids" refer to the steam-volatile neutral and acidic substances determined by the gas chromatographic method used herein.*

dentified peaks 10 and 12 than the aroma-deficient samples. Also, the three aromatic samples had a higher proportion of isovaleric to n-valeric acid than the aroma-deficient samples. However, in total acids, Aroma-deficient B had a higher level than two of the aromatic samples.

Table 2 gives a comparison of the relative amounts of total acids for both leaf and smoke. The leaf data are from a previous study and are based on total peak area (EPA) rather than amounts; however, as indicated previously (Schmeltz *et al*, 1963*b*), peak area and amount are comparable for closely related tobaccos, e.g. grades of a type. Except for Aroma-deficient B, some relationship between total amounts of volatile acids in leaf and smoke is evident. However, the free acid levels in smoke **(Table 1)** are much higher than the levels of acids which distill at the natural leaf pH **(Table 2)**, which would indicate that significant amounts of free volatile acids are released or formed during burning.

Attempts to relate organoleptic properties of leaf or smoke and total amounts of volatile acids therein are summarized in **Table 3.** The panel test results on leaf aroma were based on the average ratings of two determinations made by 15 observers, and the analytical results on leaf were from a previous publication as noted above. The ratings for smoke flavor were obtained by a panel of 22-25 smokers. The value 1-2 assigned to two samples indicates that the two samples were indistinguishable in organoleptic properties. All aroma-deficient samples were lower in leaf acids than the aromatic. Although the differences between certain samples were small, a tendency for an aroma-volatile acid relationship was observed. No consistent tendency was found for smoke flavor: Aroma-deficient B was entirely out of line.

Perhaps these findings are not unexpected. Leaf aroma and smoke flavor may be interrelated but are actually different organoleptic properties. Flavor is believed to be influenced by an optimal balance of acidic and basic constitutents. Determinations of total alkaloids and total volatile bases have shown that Aroma-deficient B tobacco is very low in those components. The smoke flavor deficiency of this tobacco may be a reflection of a deficiency in bases regardless of the acid level. Since cigarette tobacco leaves con-

Table 1. Quantitative differences in certain volatile acids of smoke condensates of cigarettes made from aromatic or aroma-deficient bright tobaccos.

Peak	Mg per 100 cigarettes*	Relative amounts** Aromatic cigarettes A	B	C	Aroma-deficient cigarettes A	B
Acetic	72.2	1.0	.65	.82	.67	.90
Propionic	15.1	1.0	.70	.79	.60	1.03
Isobutyric	2.5	1.0	.58	.71	.61	.70
n-Butyric	6.8	1.0	.74	.75	.69	.84
Isovaleric	4.0	1.0	.77	.86	.45	.68
Peaks 5a, 5b	2.8	1.0	.96	.85	1.33	.79
n-Valeric	1.7	1.0	.77	.79	.67	1.04
Peak 6a	0.3	1.0	.90	1.41	.59	1.14
β-Methylvaleric	1.9	1.0	.92	1.01	.69	.93
Isocaproic	0.7	1.0	1.40	1.73	.66	1.42
Caproic	0.5	1.0	.53	.51	.69	.61
Peak 10	1.1	1.0	.73	.95	.43	.55
Peak 11	0.3	1.0	—	—	.48	1.48
Peak 12	1.0	1.0	.86	1.01	.61	.79
n-Heptylic	2.5	1.0	.67	.87	.59	.71
Peak 14	0.4	1.0	.70	1.57	.65	1.0
Total	113.8	1.0	.69	.82	.66	.90

** For Aromatic A cigarettes. Unidentified acids were calculated as n-caproic acid. One hundred cigarettes weighed 100.6-131.6 g for the five samples (range of average weights).*

*** Values for* $\left\{\frac{\text{mg in smoke of 100 cigarettes of indicated tobacco}}{\text{mg in smoke of 100 cigarettes of Aromatic A tobacco}}\right\}$.

Table 2. Relative amounts of volatile acids in aromatic grades of bright tobacco and smoke condensates thereof.

Sample	Relative amounts* Leaf	Smoke
Aromatic A	1.0	1.0
Aromatic B	.84	.69
Aromatic C	.95	.82
Aroma-deficient A	.76	.66
Aroma-deficient B	.71	.90

** Leaf data are based on C_2-C_9 acids (Schmeltz et al, 1963b). Smoke contained only traces of C_8 and C_9 acids, and values are based on acids listed in table 1. Total volatile acids in leaf (Aromatic A) were 3.4 mg/100 g leaf. See Table 1 for corresponding levels in smoke.*

Table 3. Relationship between leaf aroma or smoke flavor and volatile acid content of aromatic, bright grades, and smoke thereof.

Sample	Relative rating Leaf Aroma*	Leaf Acids**	Smoke Flavor*	Smoke Acids**
Aromatic A	1	1	1-2	1
Aromatic B	3	3	3	4
Aromatic C	2	2	1-2	3
Aroma-deficient A	4	4	4	5
Aroma-deficient B	5	5	5	2

** Degree of aroma or flavor determined by panel testing. 1=highest aroma or flavor, 1-2=2 samples were of equal flavor.*

*** Amounts of total volatile acids (see footnotes, tables 1 and 2). 1=highest amount.*

tain little or no free volatile bases at the natural pH of the leaf, the contribution of such components to leaf aroma may be nil, and the volatile acids in leaf may be of paramount importance. Studies of the relative amounts of bases in the five smoke condensates are currently underway to elucidate this point.

It was noted that Aromatic-deficient B leaves contained a higher level (24%) of reducing sugars than the other four samples (11-17%). Although Quin *et al* (1961) have shown that added sugars do not enhance the volatile acid levels of leaf on burning, their work was done with burley tobacco, which has a higher pH than bright. Therefore, Aromatic-deficient A cigarettes (11% sugar) were smoked with or without glucose added in amounts to raise the total sugar content to the same level as Aromatic-deficient B cigarettes. The total volatile acid level in the smoke of cigarettes with added glucose was found to be about 10% higher than that of control cigarettes, and much lower than Aroma-deficient B cigarettes, showing that the differences in sugar concentrations of Aroma-deficient A and B did not account entirely for the observed differences in volatile smoke acids.

At this point, it would be well to emphasize again the proper perspective of the present series of studies. As previously noted (Burdick *et al*, 1963), these studies represent a preliminary survey of superficial compositional differences, including possible quality-composition relationships. The studies possess certain limitations: known shortcomings of the analytical methods (Burdick *et al*, 1963; Burdick and Stedman, 1963; Schmeltz *et al*, 1963*b*) the subjectivity of all panel testing, the great variability in tobacco composition, and a relatively small number of test samples evaluated. However, these studies may serve to stimulate and perhaps direct more detailed investigations, which may better determine the applicability of the preliminary findings.

Summary

Certain steam-volatile acids were determined in smoke condenstates of cigarettes made from five flue-cured tobaccos having different degrees of aromaticity. The analytical results were compared with panel test findings on the relative flavor of the cigarettes. For the acids, no consistent relationship between flavor and total amounts was observed, due mainly to one sample which did not follow the pattern.

Comparison of panel test findings of the relative aromaticity and recently published data on certain volatile leaf acids of the five tobacco samples showed that a tendency toward a relationship existed between relative leaf aroma and levels of certain volatile acids. Limitations of the findings were discussed.

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